

## **Supplementary Information**

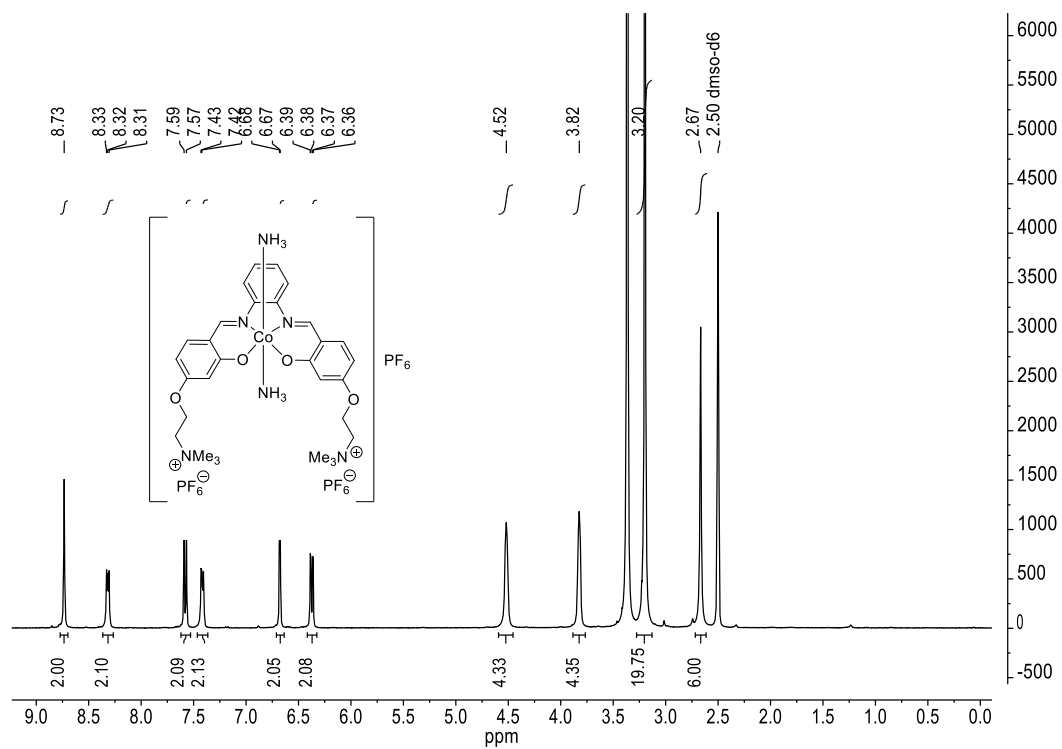


Figure S1 – <sup>1</sup>H-NMR spectrum for compound **1** in DMSO-d<sup>6</sup>.

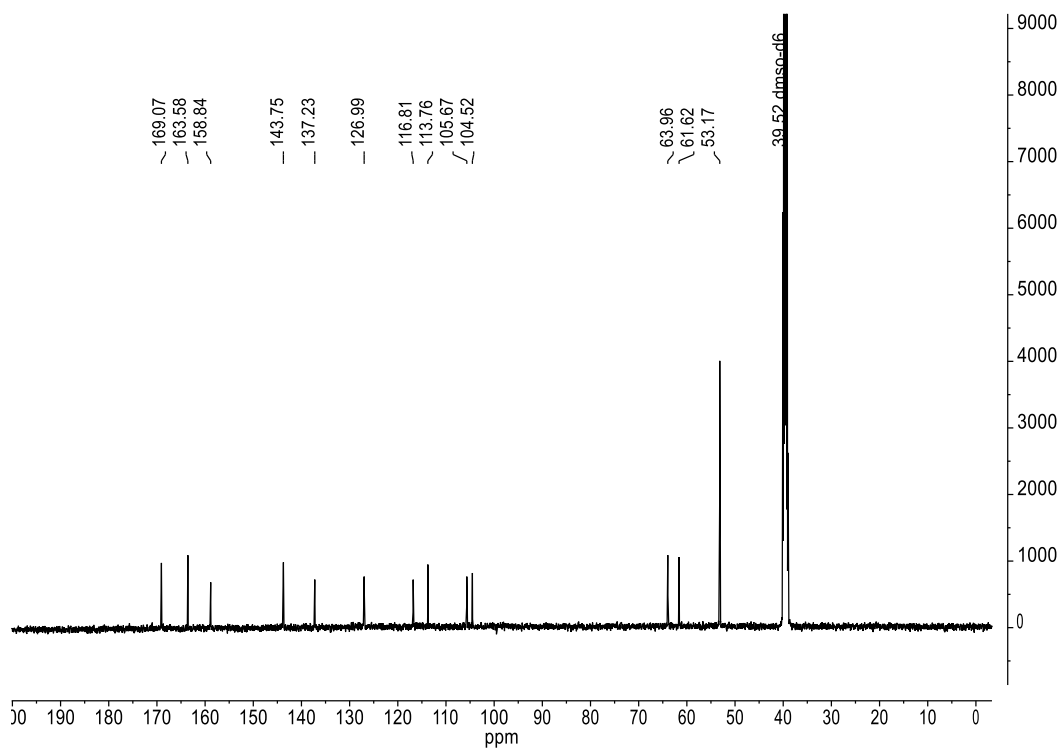


Figure S2 – <sup>13</sup>C-NMR spectrum for compound **1** in DMSO-d<sup>6</sup>.

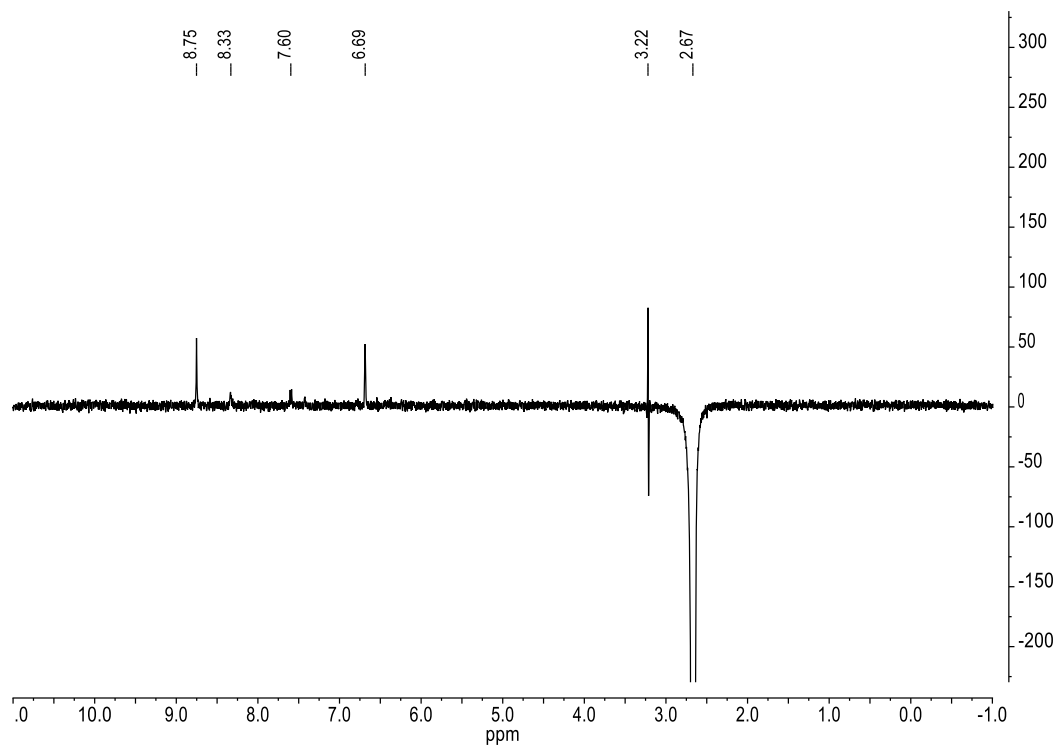


Figure S3 –  $^1\text{H}$ - $^1\text{H}$  Selective ROESY (2.67 ppm,  $\text{NH}_3$  signal) for compound **1** in  $\text{DMSO-d}^6$ .

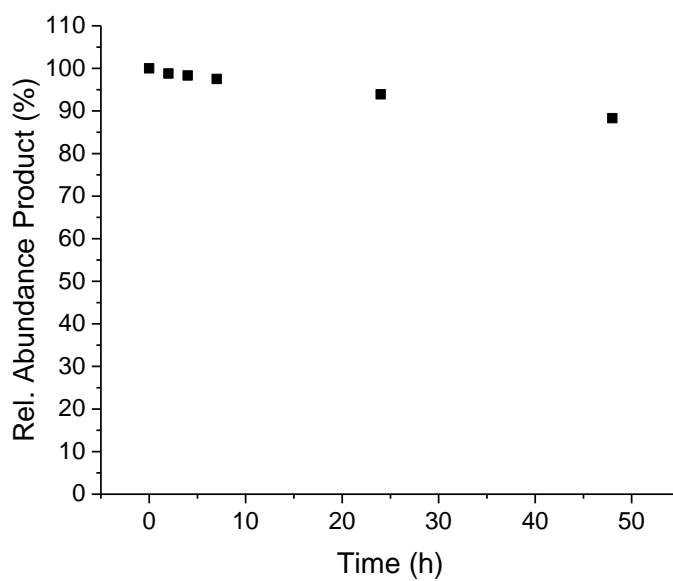
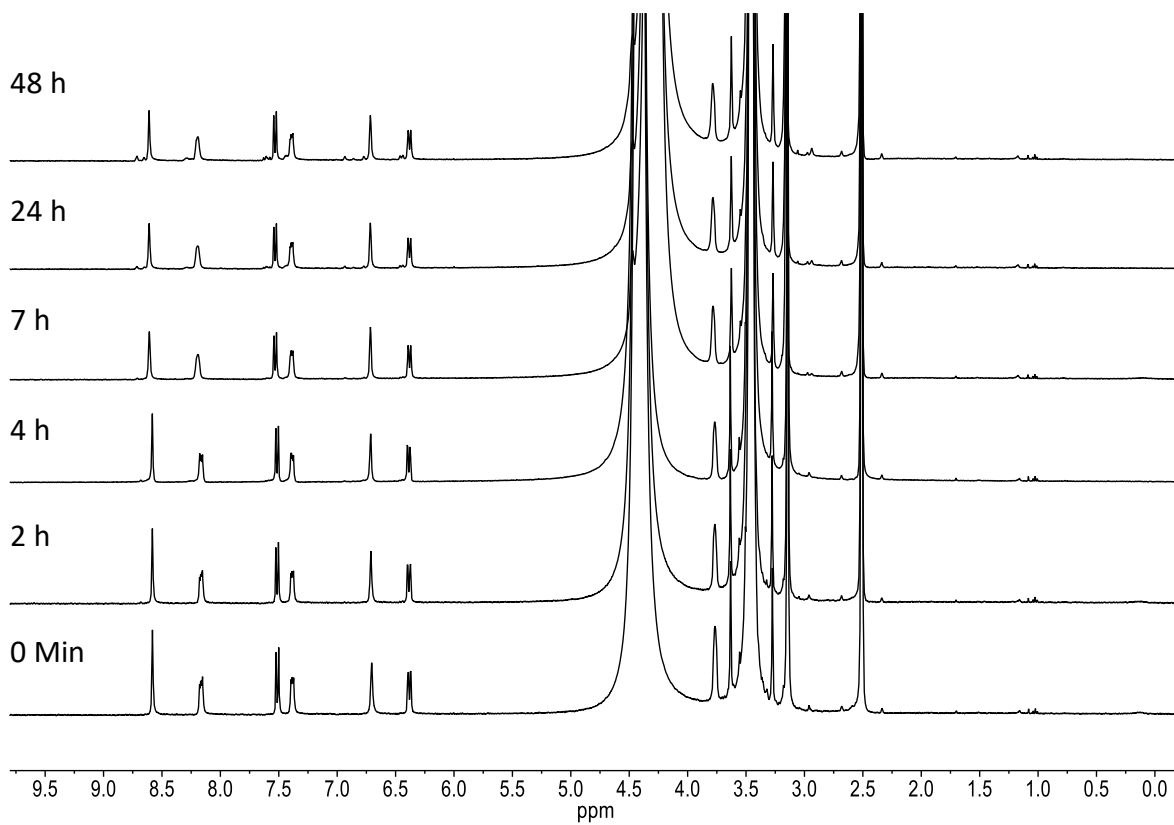


Figure S4 – Time-dependent <sup>1</sup>H-NMR spectroscopy for compound **1** in 1 M TRIS – 100 mM KCl (pH 7.2, prepared in D<sub>2</sub>O/H<sub>2</sub>O (1:9)) / DMSO (1:1).

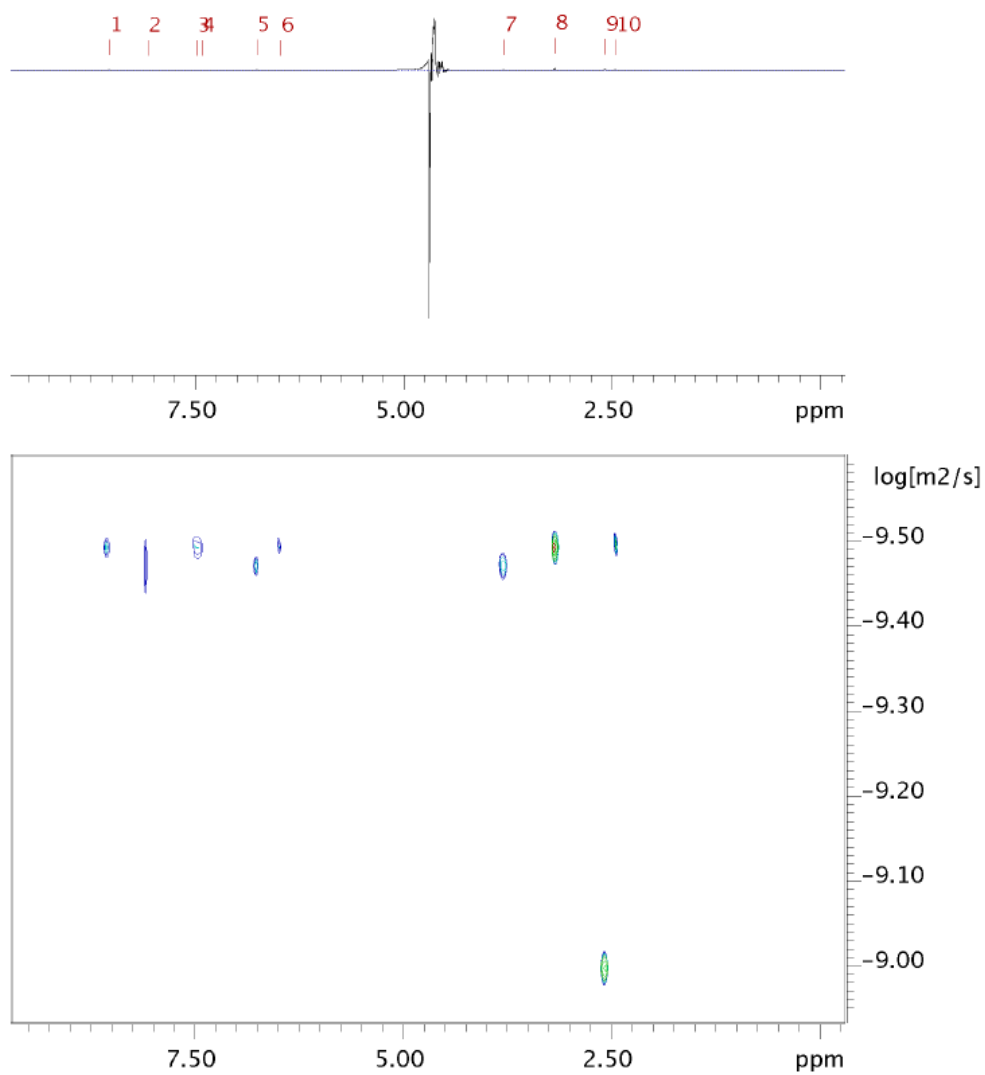


Figure S5 – Time-dependent Diffusion Ordered Spectroscopy (DOSY) for compound **1** in D<sub>2</sub>O/H<sub>2</sub>O (1:9) after 0 Min (signal at -9.00 m<sup>2</sup>s<sup>-1</sup> was assigned to free DMSO added for solubility purposes).

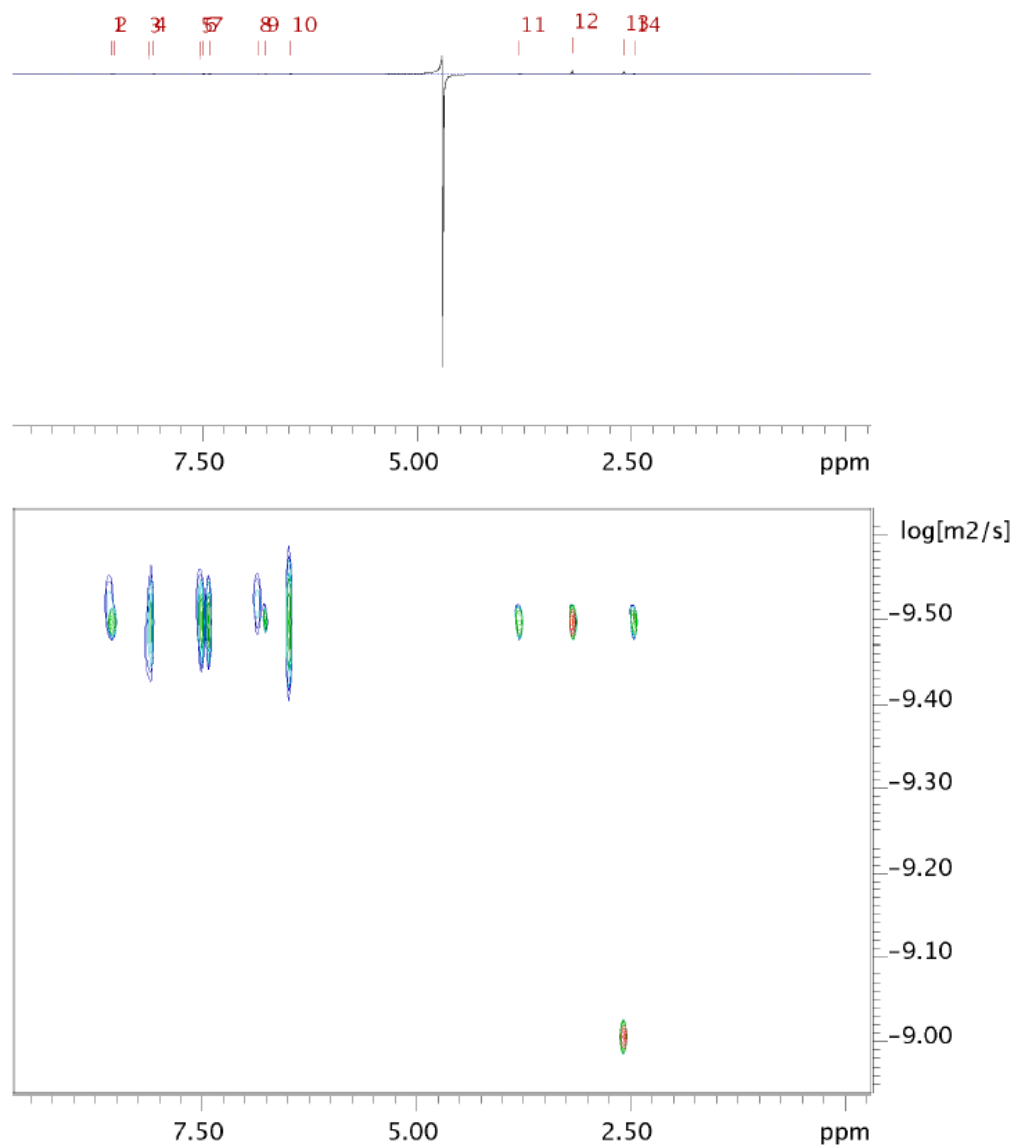


Figure S6 – Time-dependent Diffusion Ordered Spectroscopy (DOSY) for compound **1** in D<sub>2</sub>O/H<sub>2</sub>O (1:9) after 24 h (signal at -9.00 m<sup>2</sup>s<sup>-1</sup> was assigned to free DMSO added for solubility purposes).

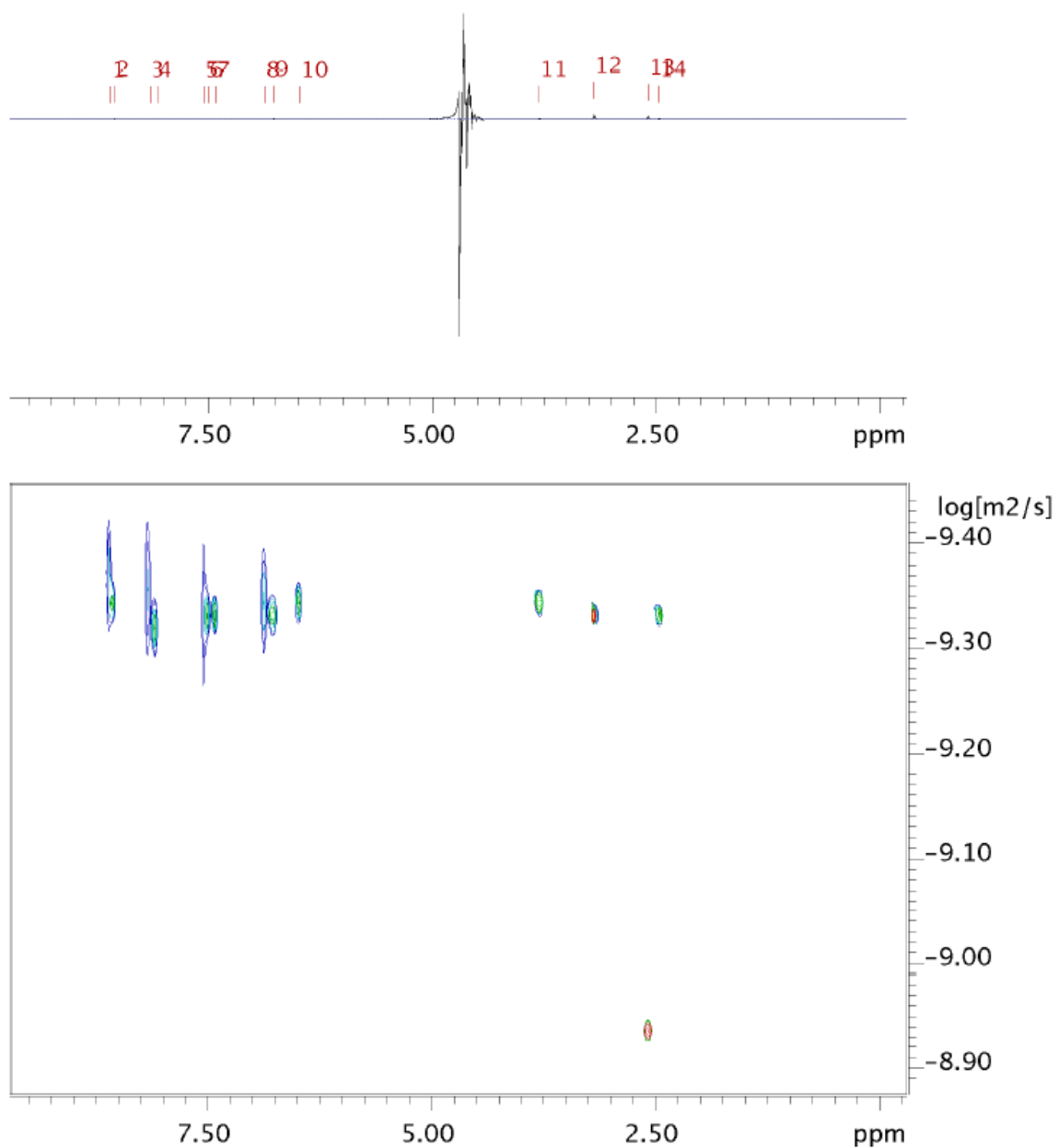


Figure S7 – Time-dependent Diffusion Ordered Spectroscopy (DOSY) for compound **1** in D<sub>2</sub>O/H<sub>2</sub>O (1:9) after 48 h (signal at -9.00 m<sup>2</sup>s<sup>-1</sup> was assigned to free DMSO added for solubility purposes).

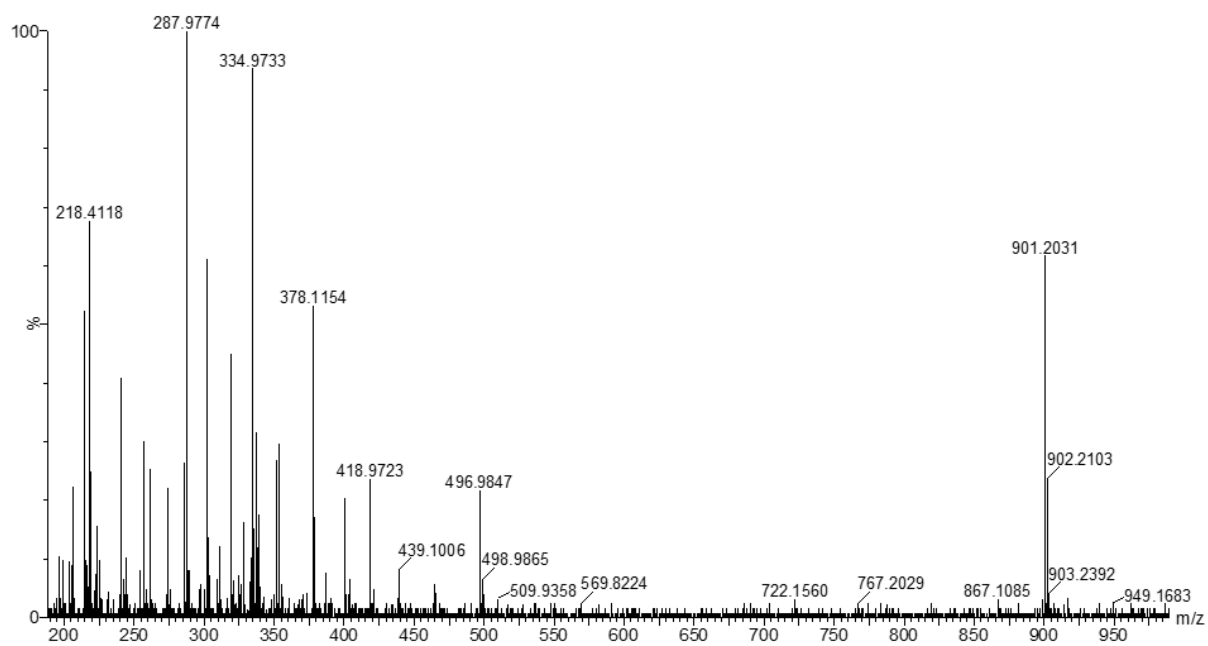


Figure S8 – ESI(+) mass spectrum for compound 1.



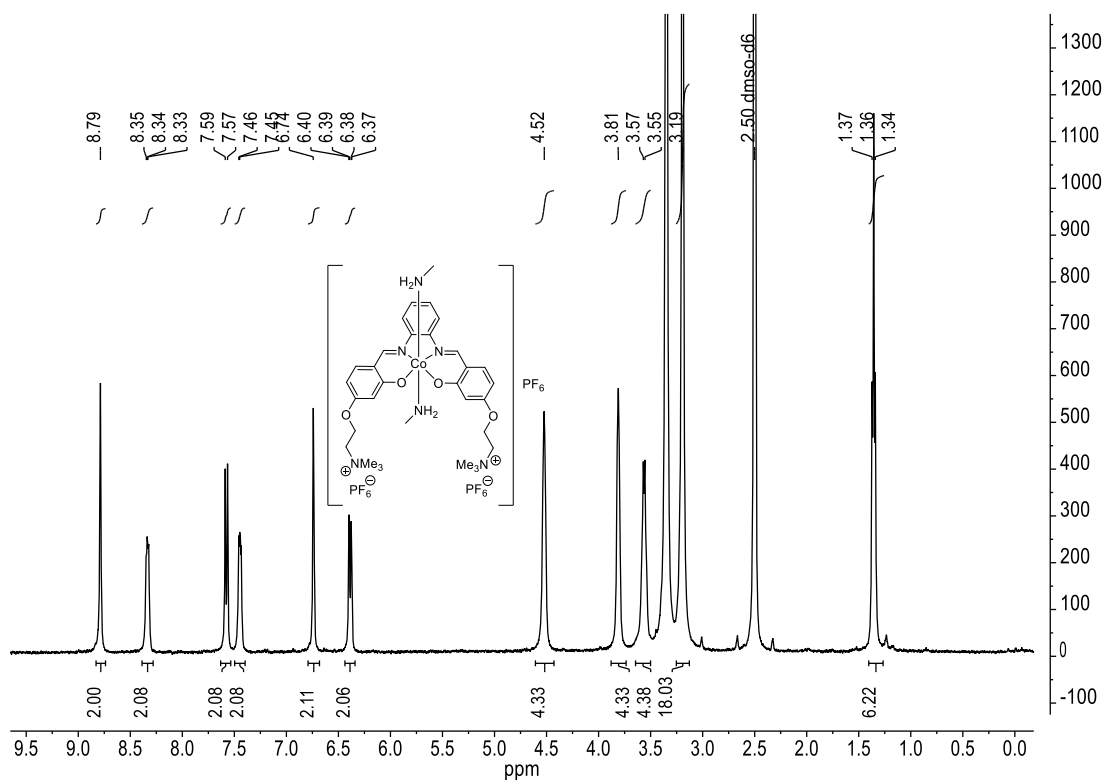


Figure S9 – <sup>1</sup>H-NMR spectrum for compound 2 in DMSO-d<sup>6</sup>.

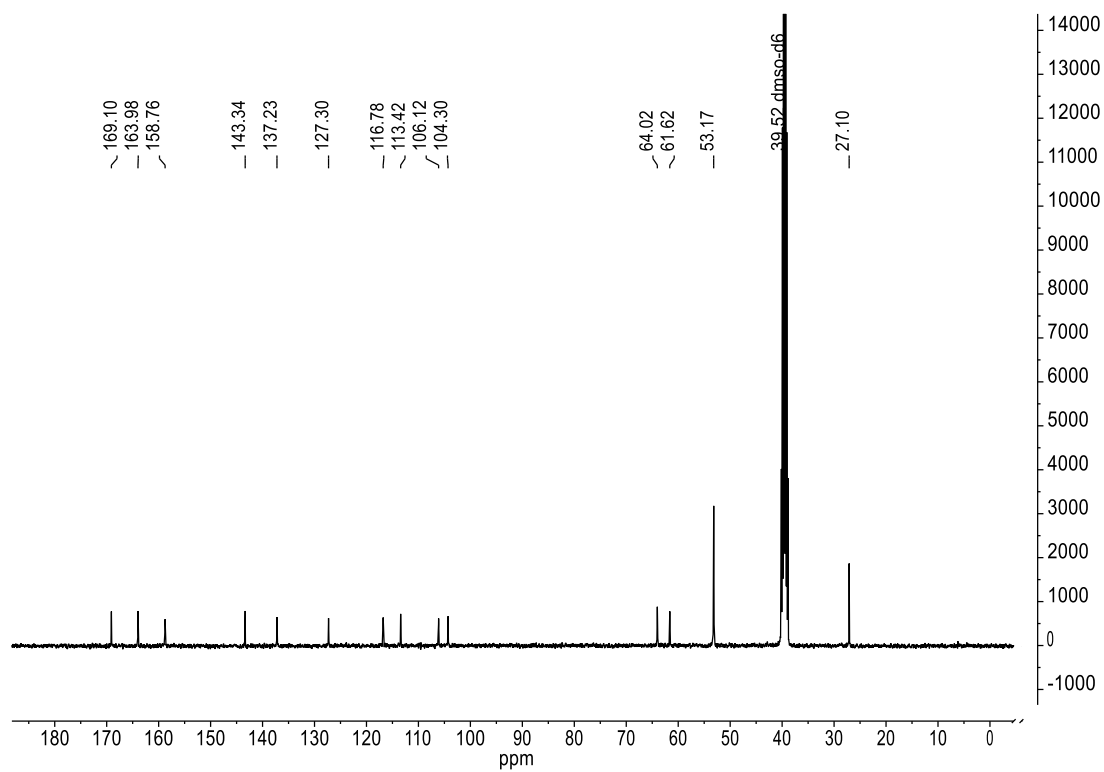


Figure S10 – <sup>13</sup>C-NMR spectrum for compound 2 in DMSO-d<sup>6</sup>.

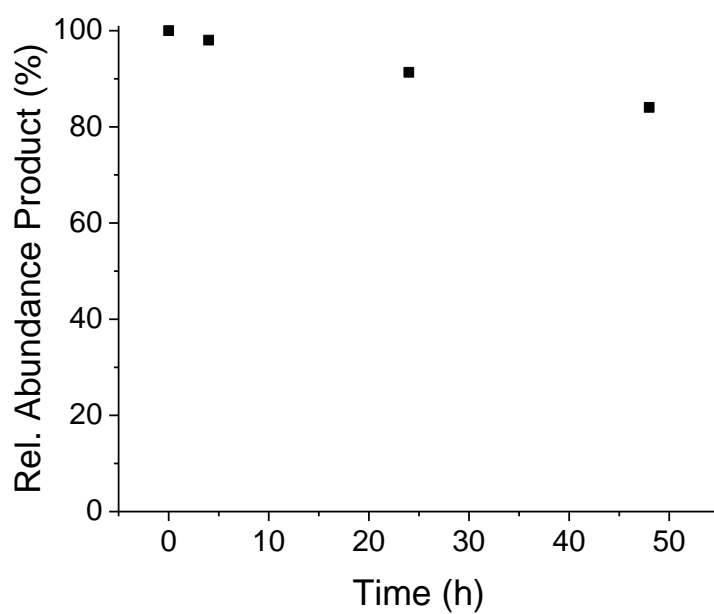
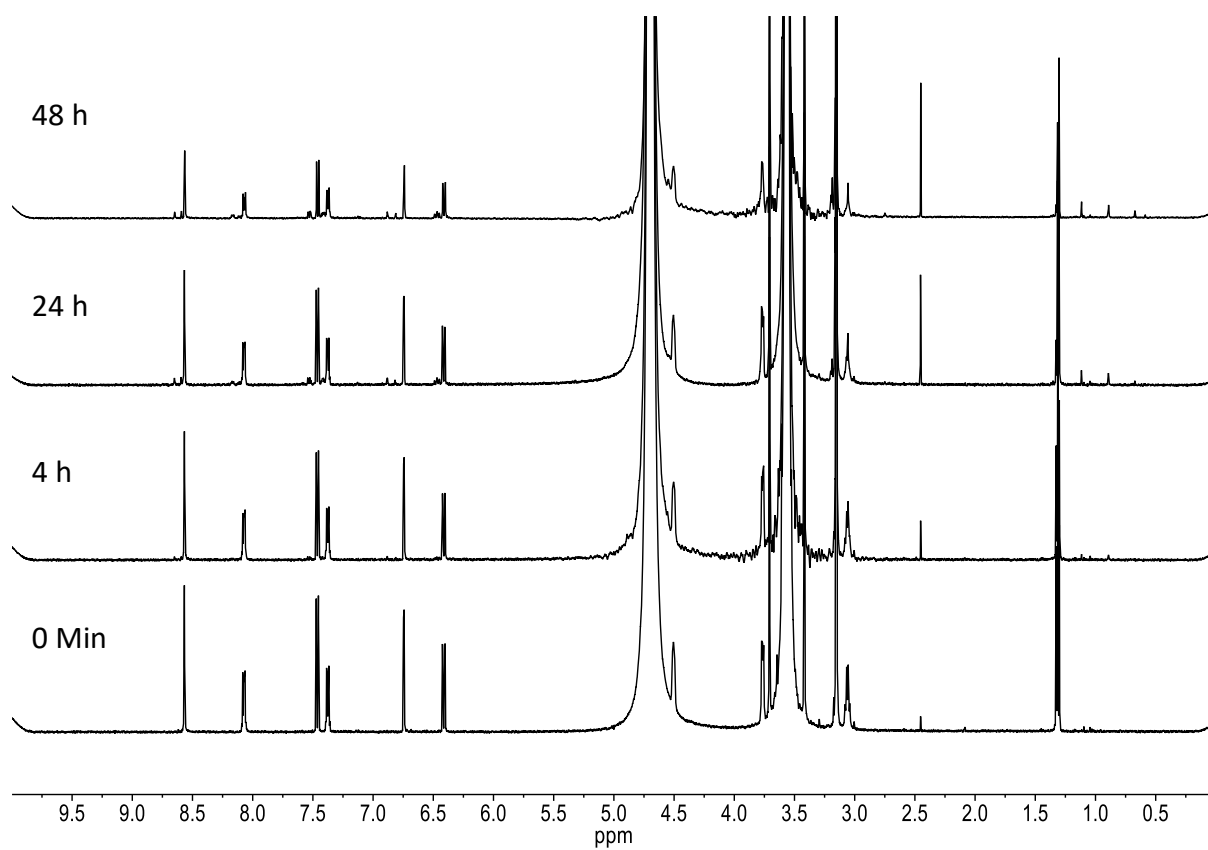


Figure S11 – Time-dependent <sup>1</sup>H-NMR spectroscopy for compound 2 in 1 M TRIS – 100 mM KCl (pH 7.2, prepared in D<sub>2</sub>O/H<sub>2</sub>O (1:9)).

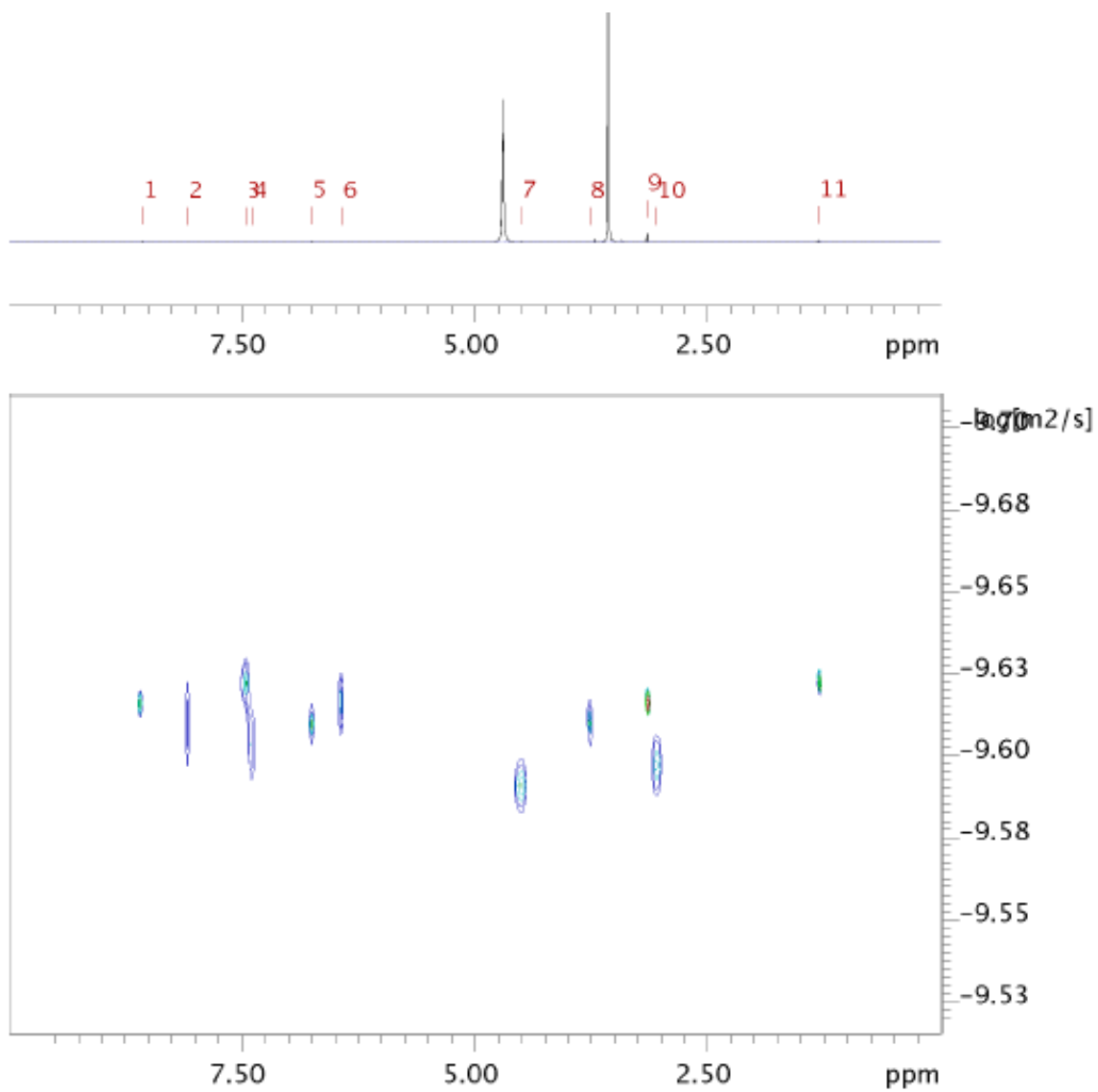


Figure S12 – Time-dependent Diffusion Ordered Spectroscopy (DOSY) after 0 Min for compound **2** in 1 M TRIS – 100 mM KCl (pH 7.2, prepared in D<sub>2</sub>O/H<sub>2</sub>O (1:9)).

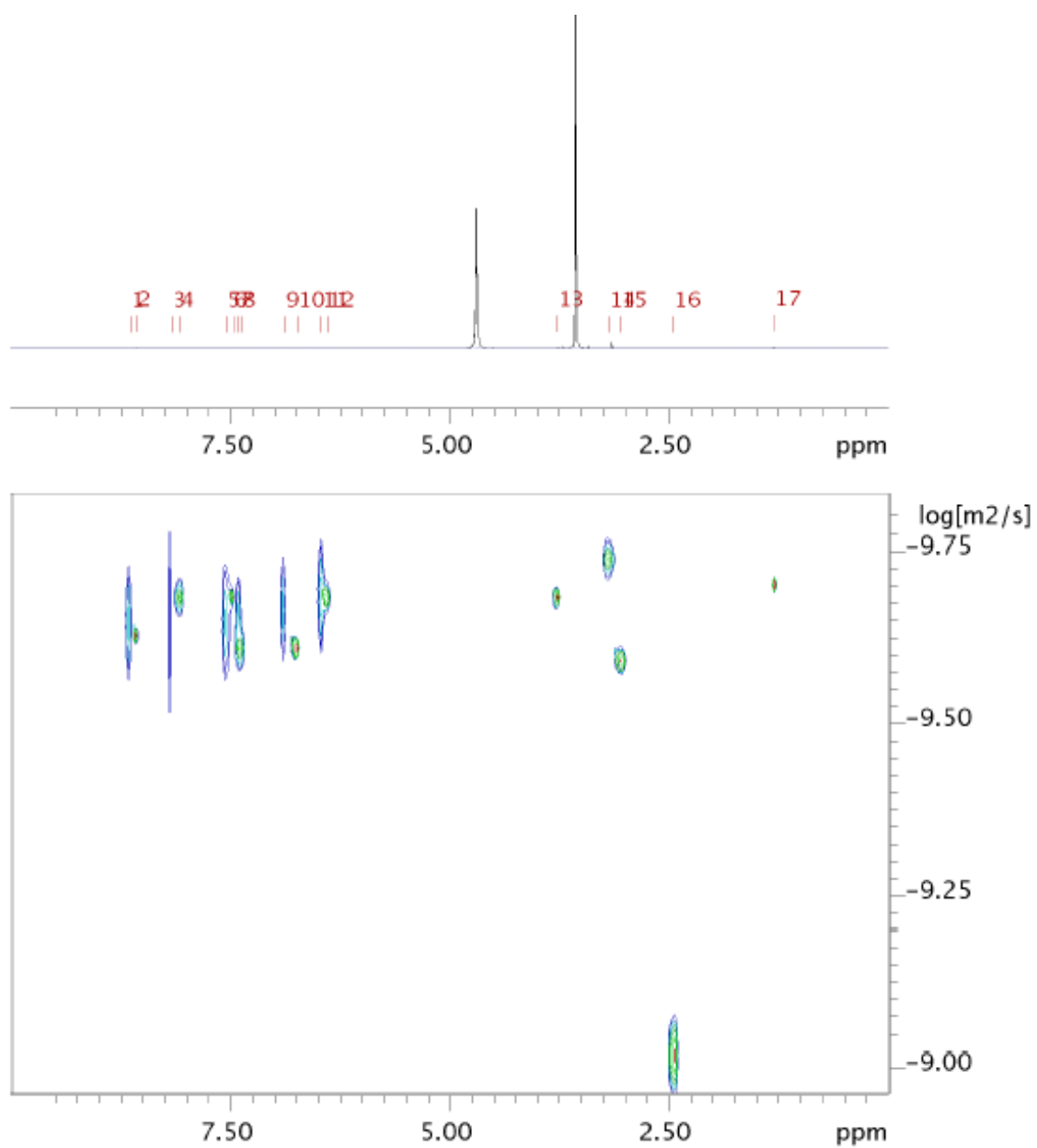


Figure S13 – Time-dependent Diffusion Ordered Spectroscopy (DOSY) after 24 h for compound **2** in 1 M TRIS – 100 mM KCl (pH 7.2, prepared in D<sub>2</sub>O/H<sub>2</sub>O (1:9)).

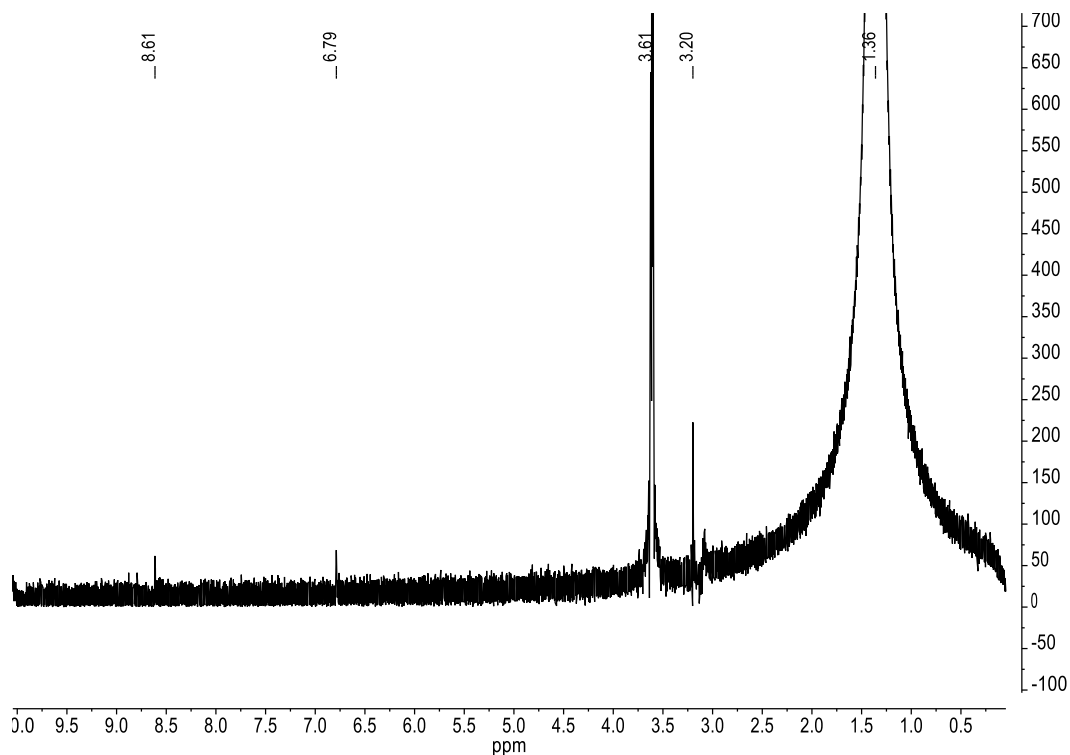


Figure S14 –  $^1\text{H}$ - $^1\text{H}$  Selective ROESY (1.36 ppm,  $\text{CH}_3$  signal) after 0 Min for compound **2** in 1 M TRIS – 100 mM KCl (pH 7.2, prepared in  $\text{D}_2\text{O}/\text{H}_2\text{O}$  (1:9)).

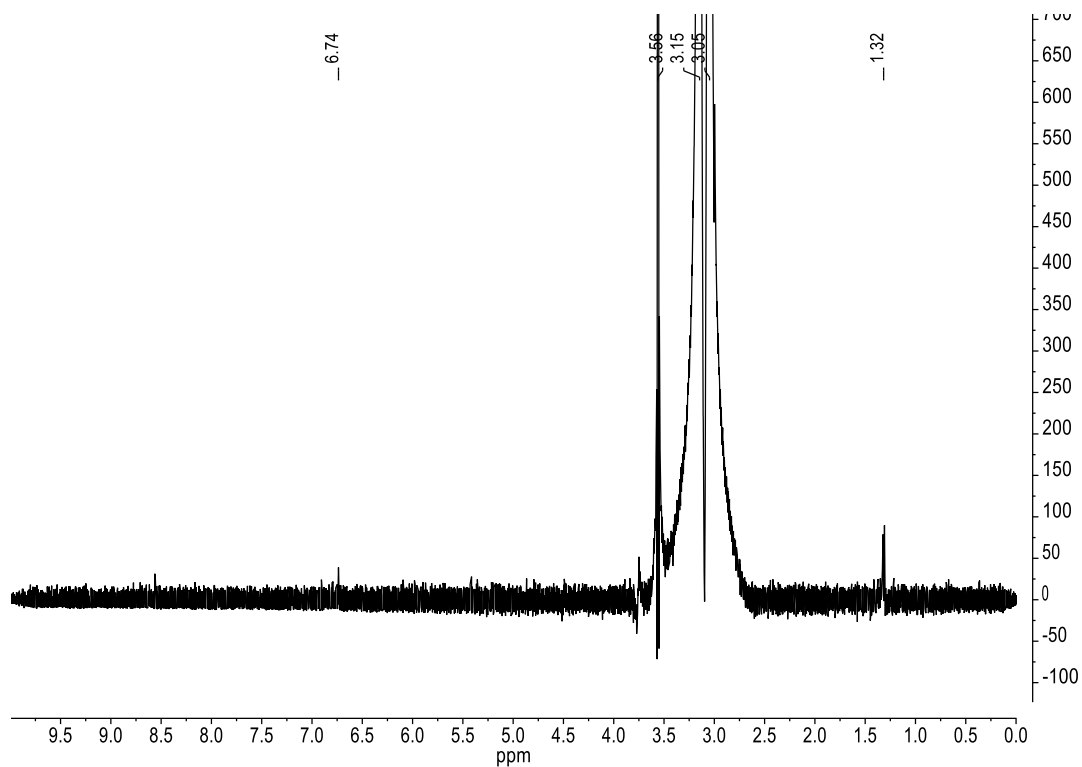


Figure S15 –  $^1\text{H}$ - $^1\text{H}$  Selective ROESY (3.56 ppm,  $\text{NH}_2$  signal) after 0 Min for compound **2** in 1 M TRIS – 100 mM KCl (pH 7.2, prepared in  $\text{D}_2\text{O}/\text{H}_2\text{O}$  (1:9)).

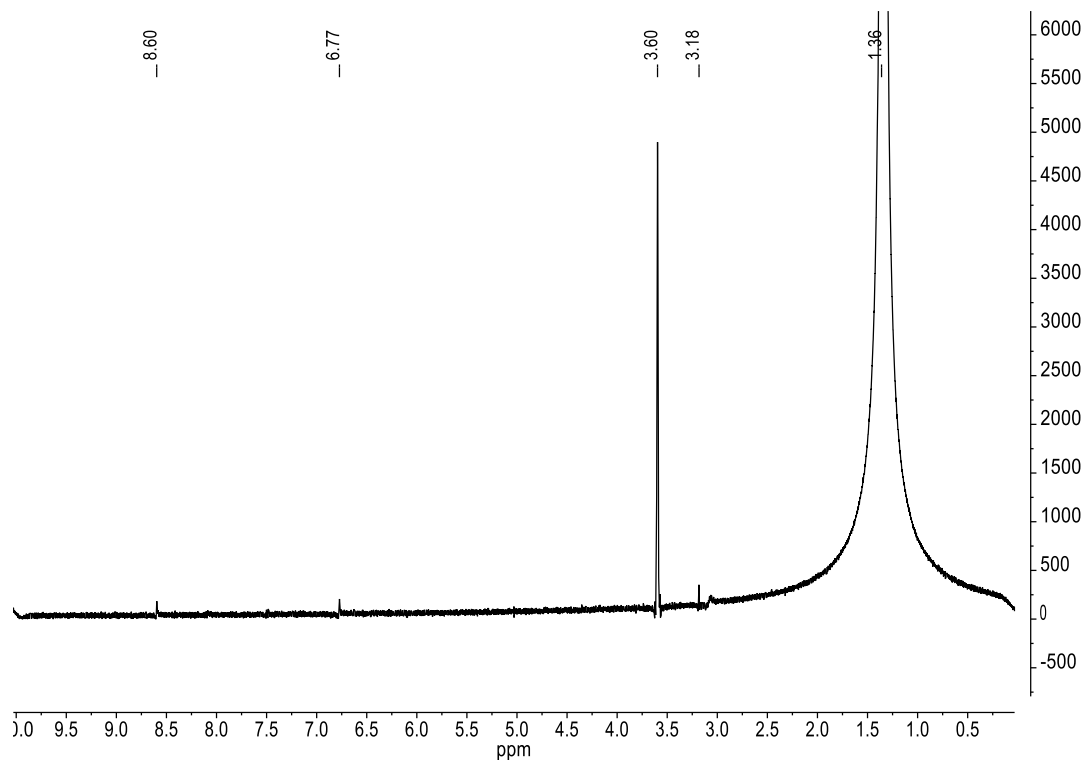


Figure S16 –  $^1\text{H}$ - $^1\text{H}$  Selective ROESY (1.36 ppm,  $\text{CH}_3$  signal) after 24 h for compound **2** in 1 M TRIS – 100 mM KCl (pH 7.2, prepared in  $\text{D}_2\text{O}/\text{H}_2\text{O}$  (1:9)).

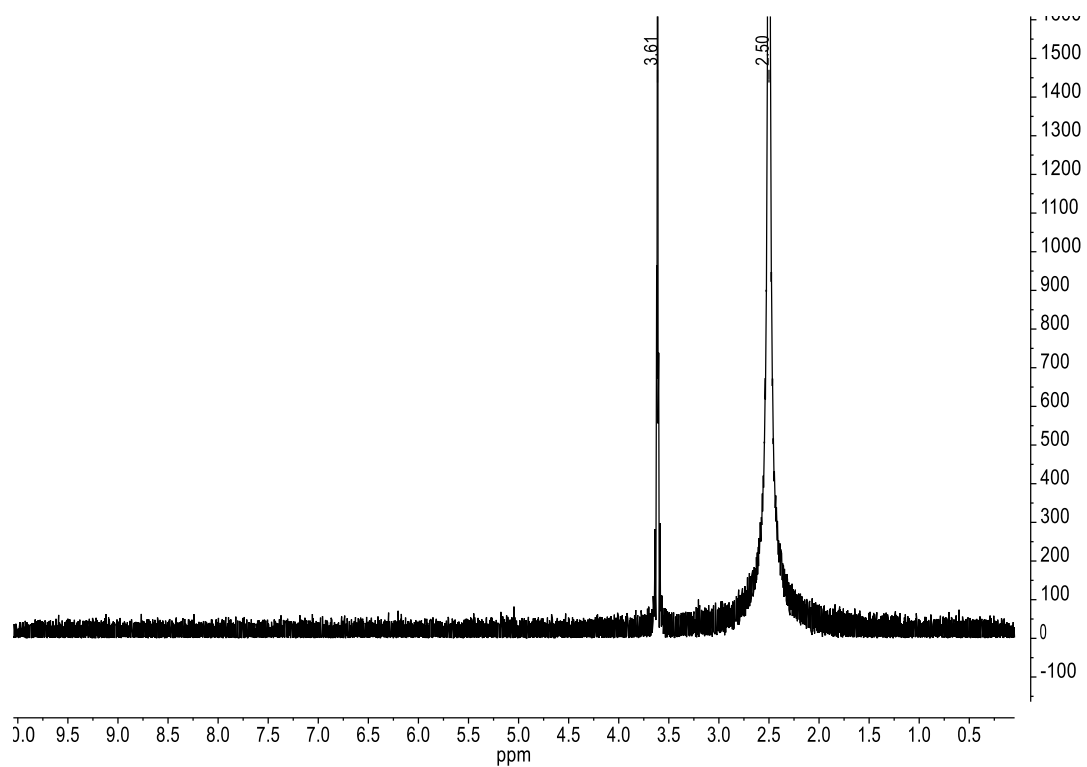


Figure S17 –  $^1\text{H}$ - $^1\text{H}$  Selective ROESY (2.50 ppm, free  $\text{NH}_2\text{Me}$ ) after 24 h for compound **2** in 1 M TRIS – 100 mM KCl (pH 7.2, prepared in  $\text{D}_2\text{O}/\text{H}_2\text{O}$  (1:9)).

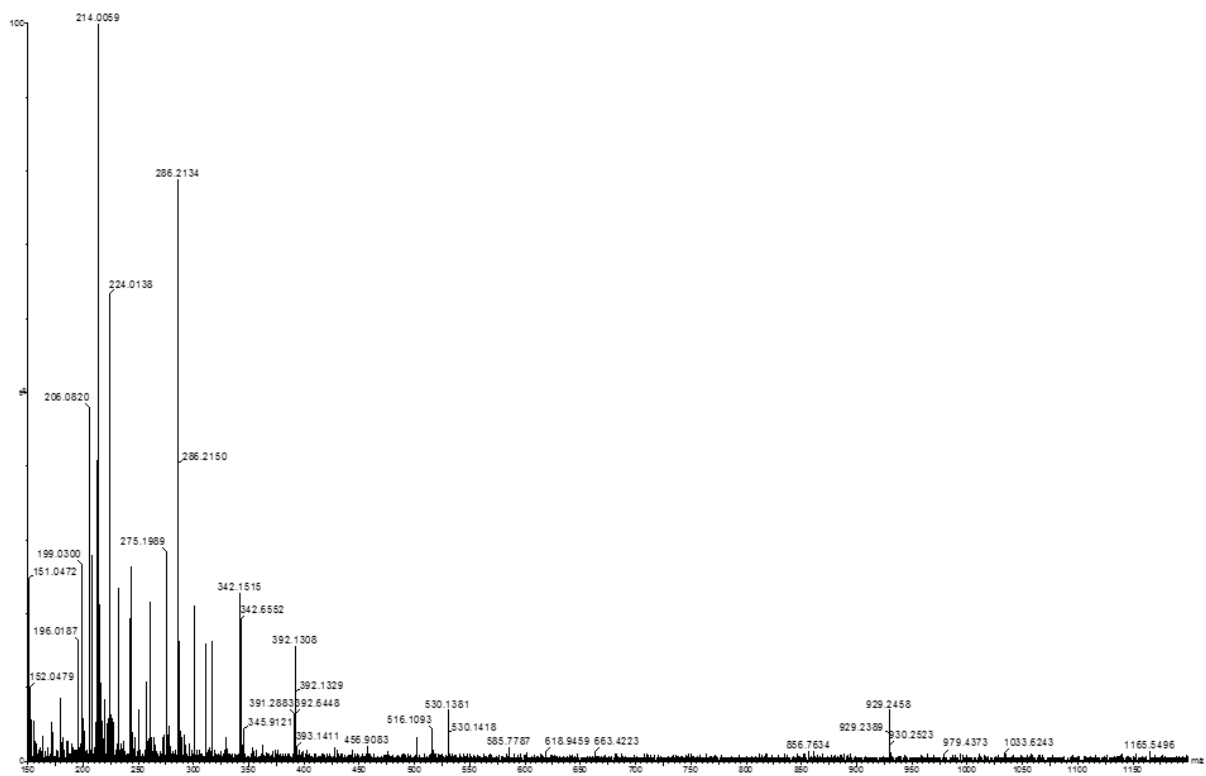


Figure S18 – ESI(+) mass spectrum for compound 2.

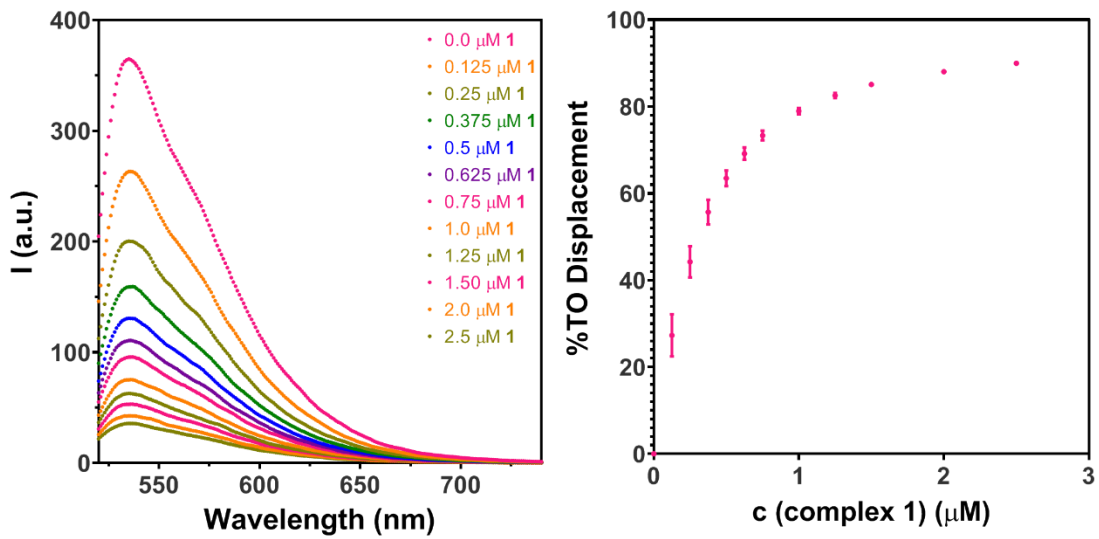


Figure S19 – FID results for titration of 0.25  $\mu\text{M}$  *HTelo* (K) and 0.50  $\mu\text{M}$  TO with increasing amounts of complex 1 (0 – 2.5  $\mu\text{M}$ ): emission spectra (left) and %TO displacement (right). Titration was performed in 10 mM Licac + 100 mM KCl buffer (pH 7.2) at rt.

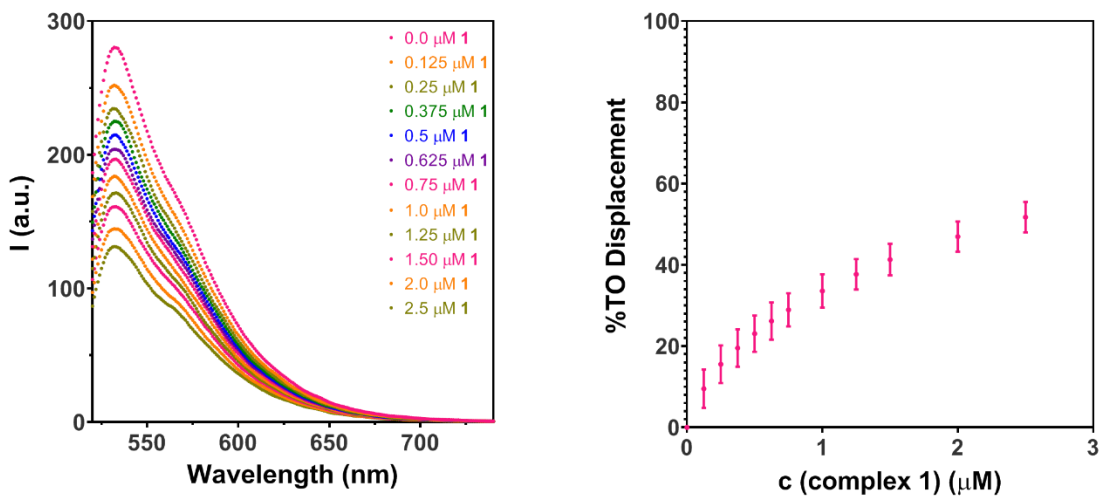


Figure S20 – FID results for titration of 0.25  $\mu\text{M}$  *HTelo* (Na) and 0.50  $\mu\text{M}$  TO with increasing amounts of complex 1 (0 – 2.5  $\mu\text{M}$ ): emission spectra (left) and %TO displacement (right). Titration was performed in 10 mM Licac + 100 mM KCl buffer (pH 7.2) at rt.



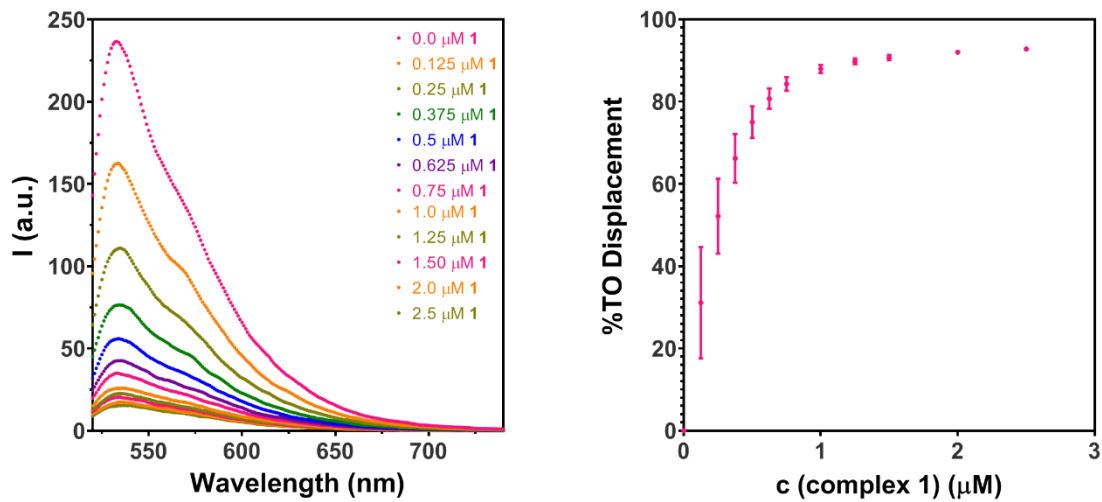


Figure S21 – FID results for titration of 0.25 μM *c-myc* and 0.50 μM TO with increasing amounts of complex 1 (0 – 2.5 μM): emission spectra (left) and %TO displacement (right). Titration was performed in 10 mM Licac + 100 mM KCl buffer (pH 7.2) at rt.

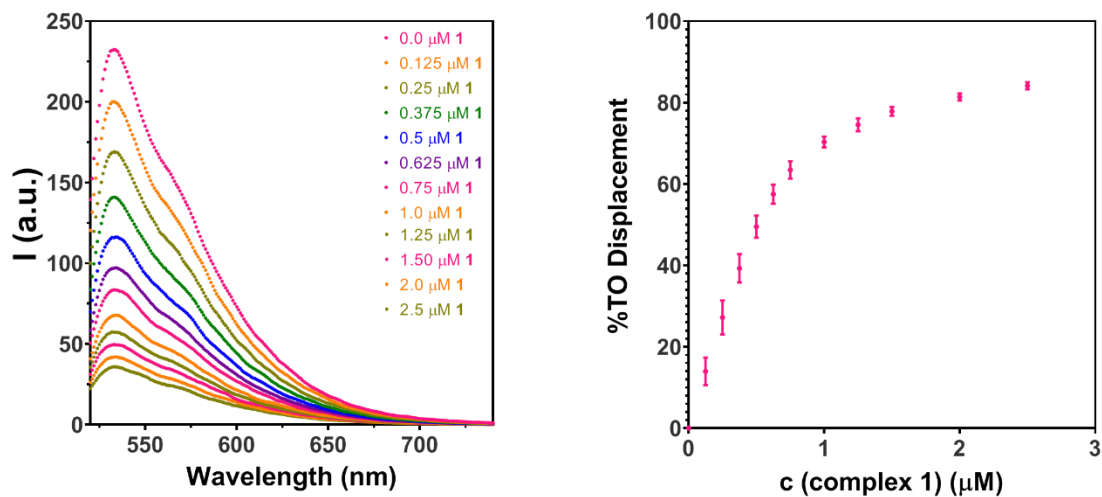


Figure S22 – FID results for titration of 0.25 μM *c-kit2* and 0.50 μM TO with increasing amounts of complex 1 (0 – 2.5 μM): emission spectra (left) and %TO displacement (right). Titration was performed in 10 mM Licac + 100 mM KCl buffer (pH 7.2) at rt.

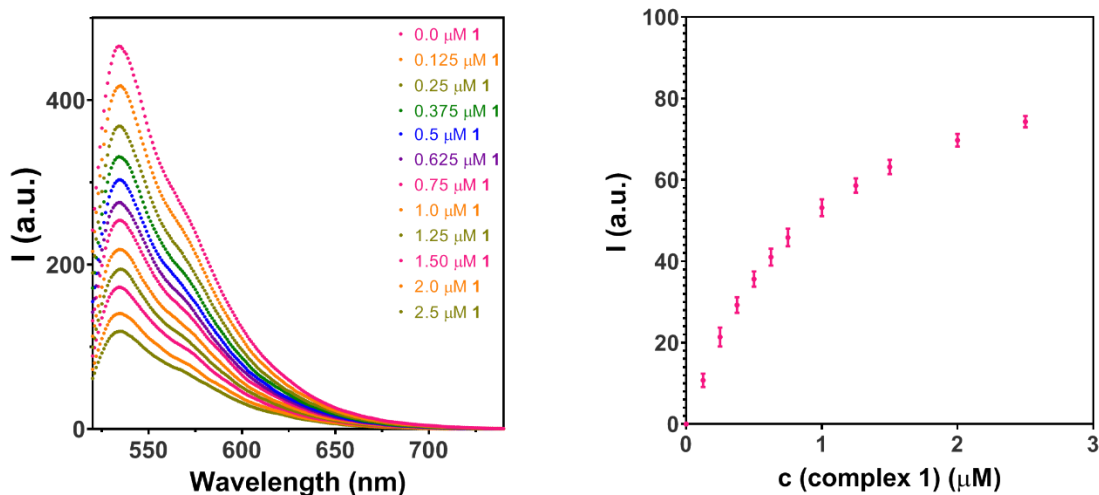


Figure S23 – FID results for titration of 0.25  $\mu\text{M}$  *bcl2* and 0.50  $\mu\text{M}$  TO with increasing amounts of complex **1** (0 – 2.5  $\mu\text{M}$ ): emission spectra (left) and %TO displacement (right). Titration was performed in 10 mM Licac + 100 mM KCl buffer (pH 7.2) at rt.

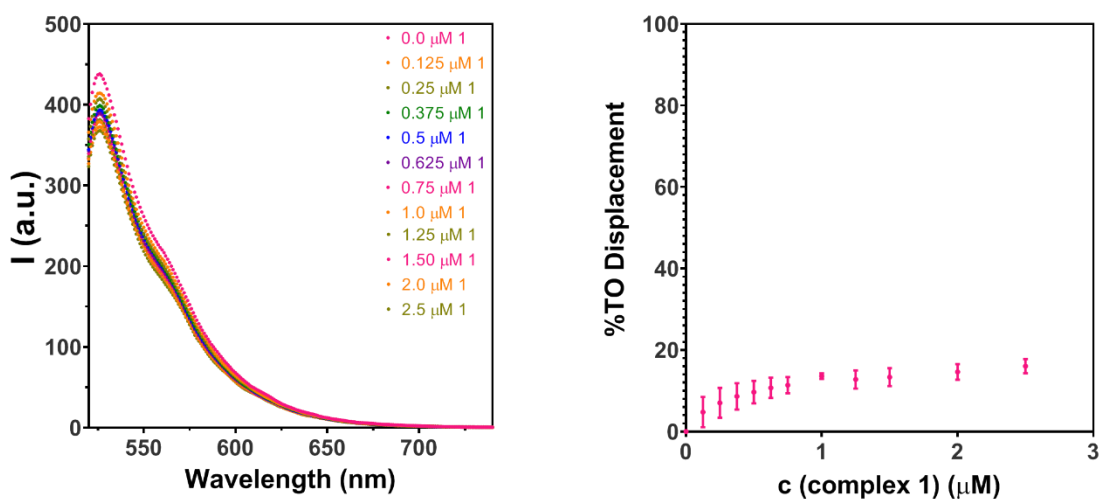


Figure S24 – FID results for titration of 0.25  $\mu\text{M}$  *ds26* and 0.75  $\mu\text{M}$  TO with increasing amounts of complex **1** (0 – 2.5  $\mu\text{M}$ ): emission spectra (left) and %TO displacement (right). Titration was performed in 10 mM Licac + 100 mM KCl buffer (pH 7.2) at rt.

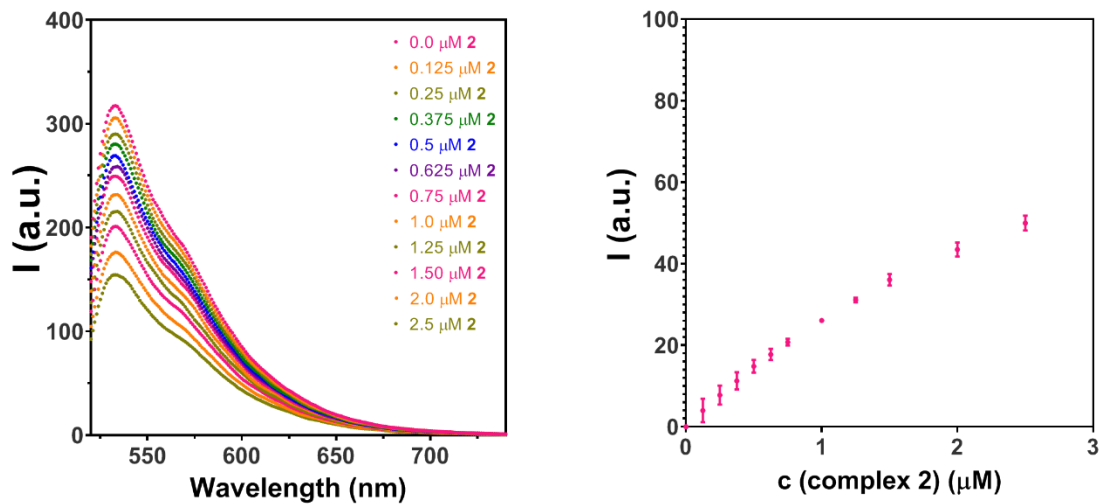


Figure S25 – FID results for titration of 0.25  $\mu\text{M}$  *c-myc* and 0.50  $\mu\text{M}$  TO with increasing amounts of complex 2 (0 – 2.5  $\mu\text{M}$ ): emission spectra (left) and %TO displacement (right). Titration was performed in 10 mM Licac + 100 mM KCl buffer (pH 7.2) at rt.

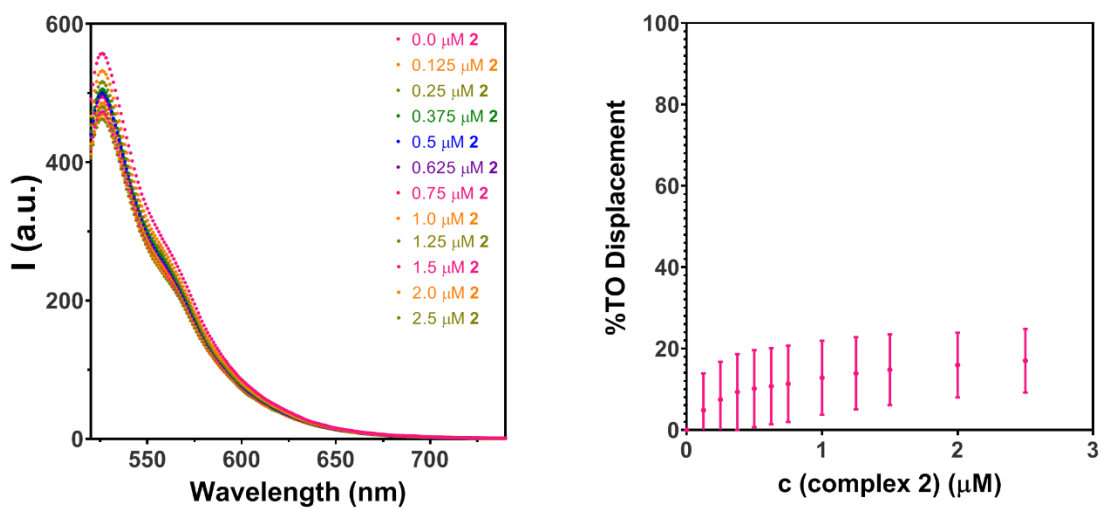


Figure S26 – FID results for titration of 0.25  $\mu\text{M}$  *ds26* and 0.75  $\mu\text{M}$  TO with increasing amounts of complex 2 (0 – 2.5  $\mu\text{M}$ ): emission spectra (left) and %TO displacement (right). Titration was performed in 10 mM Licac + 100 mM KCl buffer (pH 7.2) at rt.

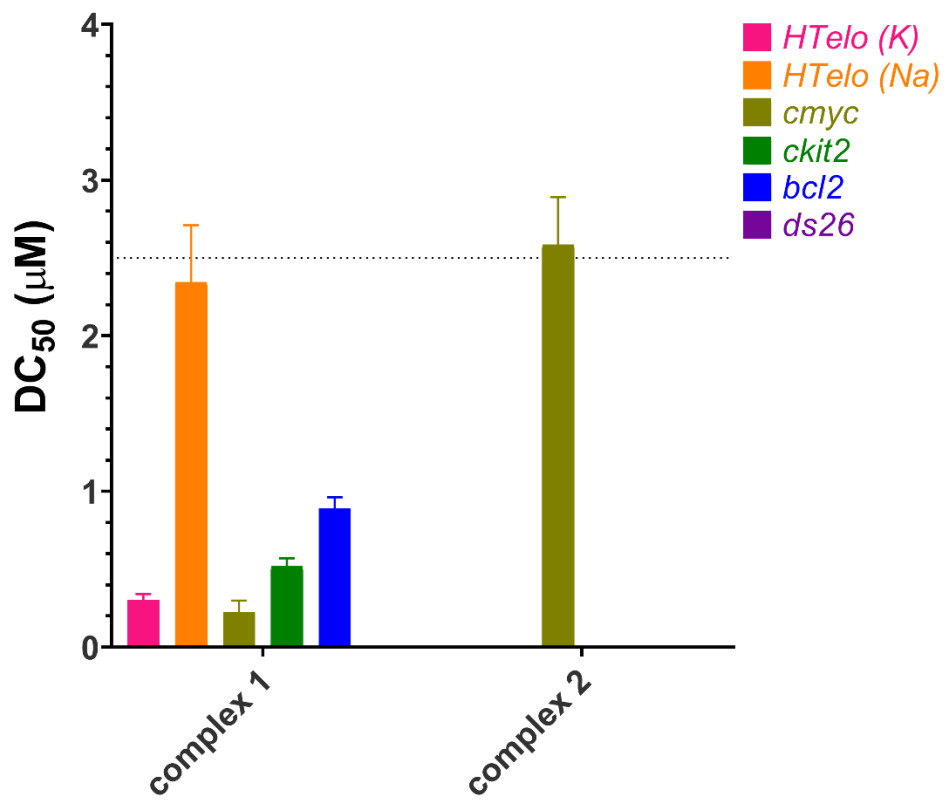


Figure S27 – DC<sub>50</sub> values for FID assays of complex 1 and 2 with different DNA sequences.

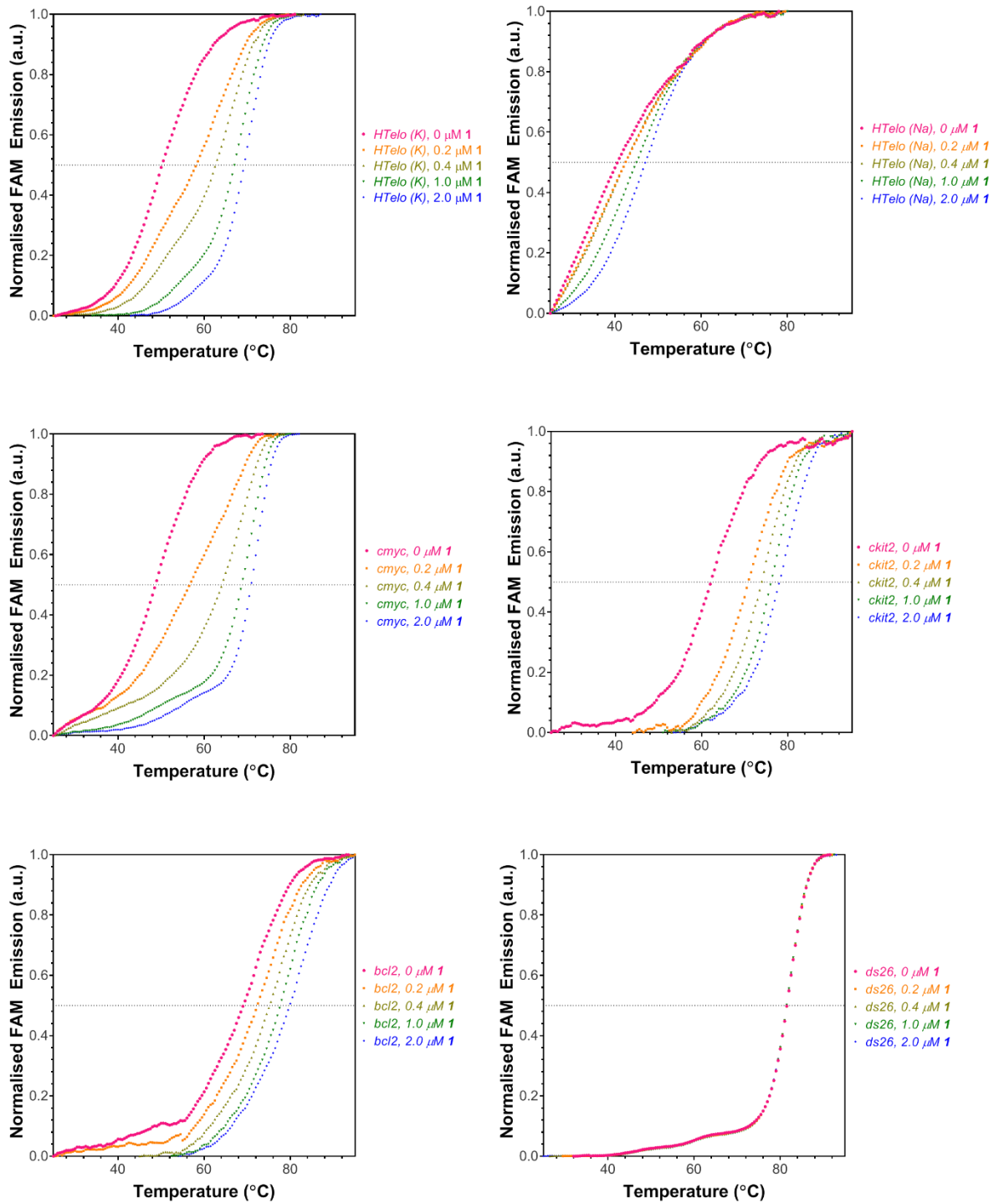


Figure S28 – FRET melting curves of different labelled DNA sequences in presence of complex 1. Melting experiments were performed in triplicate. The mean is plotted above.

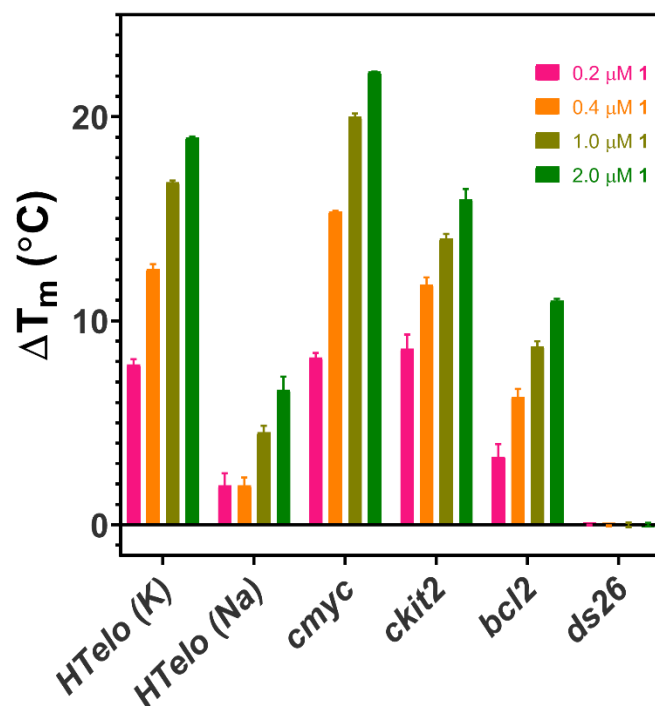


Figure S29 –  $\Delta T_m$  (°C) values for different DNA sequences are plotted in presence of increasing amounts of complex **1**. Results were obtained by averaging three independent experiments.

Table S1 –  $\Delta T_m$  (°C) values at a concentration of 1  $\mu\text{M}$  of complex **1** are listed for different DNA sequences. Triplicate experiments were used to calculate the standard deviation.

DNA sequence	$\Delta T_m$ (°C), 1 $\mu\text{M}$
<i>HTelo (K)</i>	16.8 ± 0.1
<i>HTelo (Na)</i>	4.5 ± 0.3
<i>c-myc</i>	20.0 ± 0.2
<i>c-kit2</i>	14.0 ± 0.2
<i>bcl-2</i>	8.7 ± 0.3
<i>ds26</i>	0.0 ± 0.1

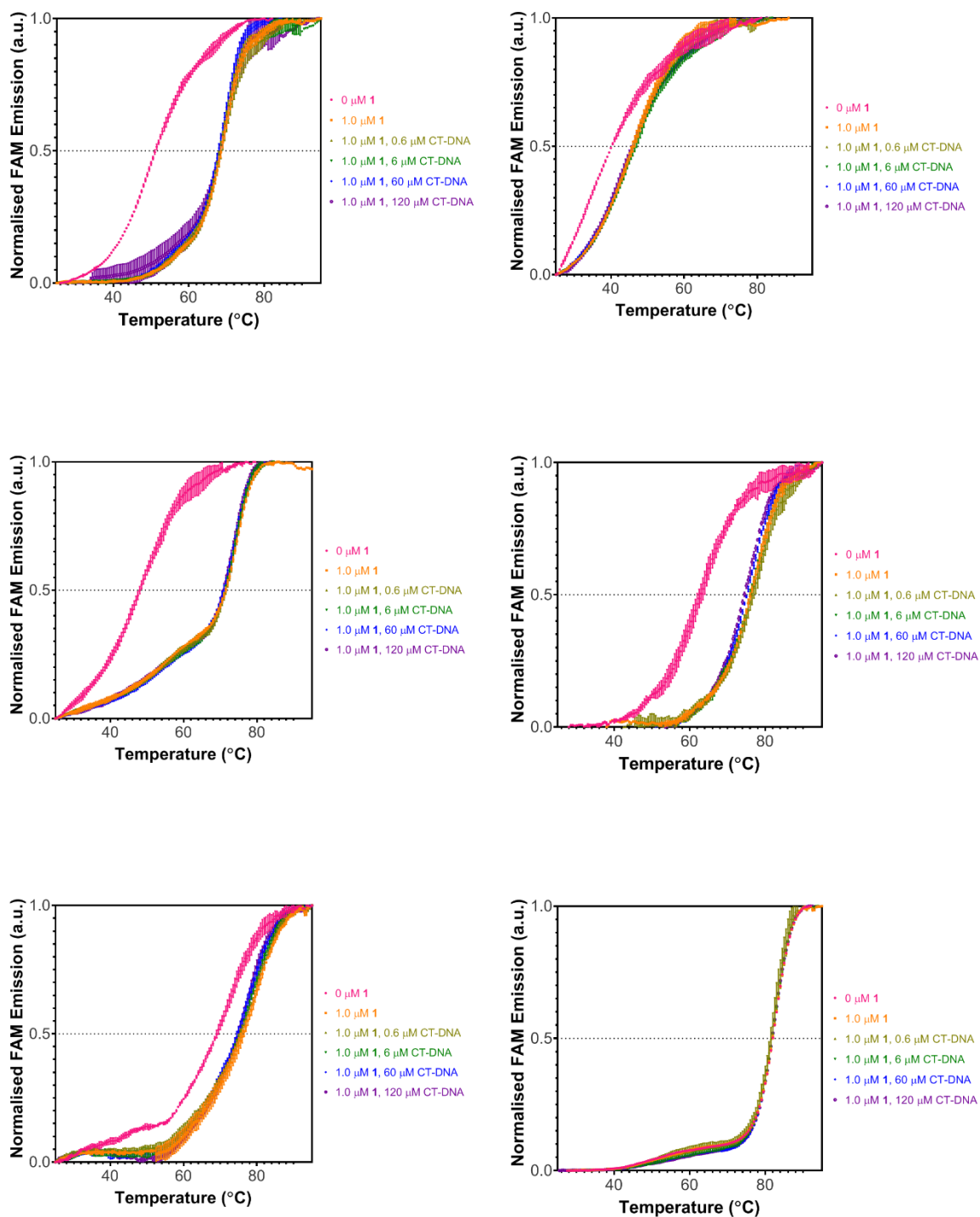


Figure S30 – FRET melting curves obtained for the competition assay are plotted above for a number of different DNA sequences in presence of complex **1**. Up to 600  $\mu\text{M}$  CT-DNA were added as competitor DNA. Details to conditions used for the experiments see Experimental Details. The mean is plotted for triplicate experiments.

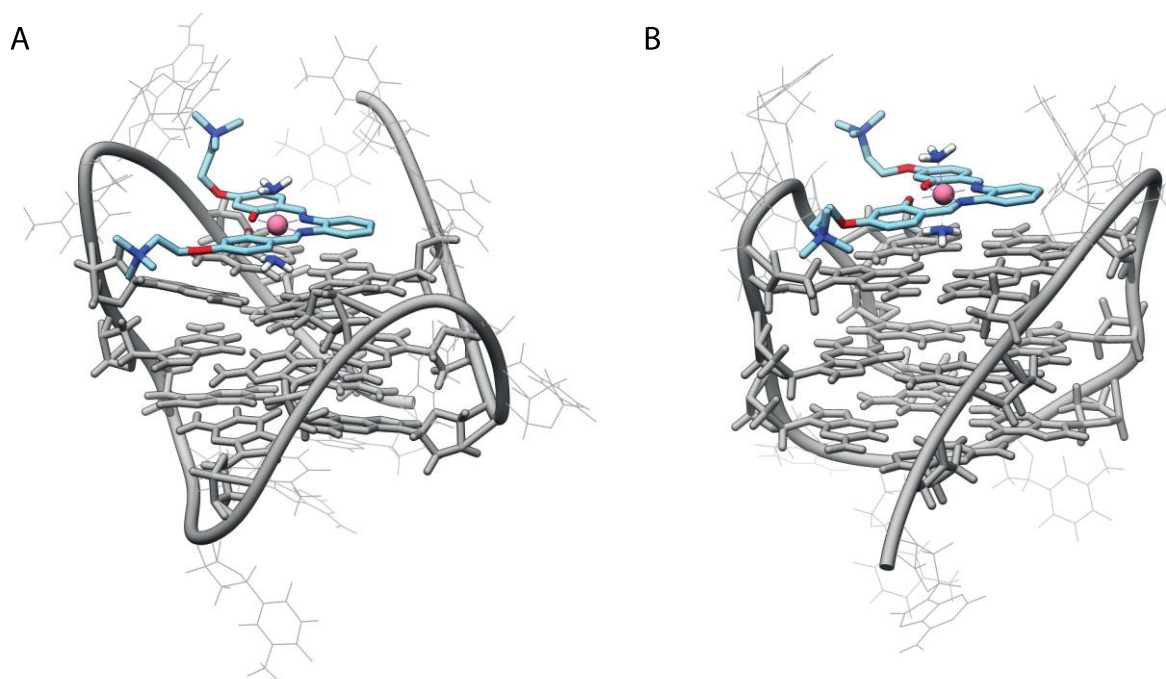
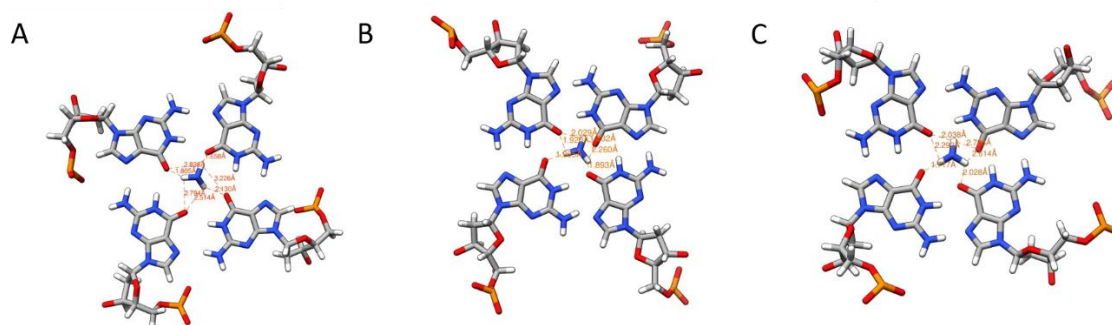


Figure S31 – Molecular docking results for compound **1** with HTelo hybrid type (PDB: 2mb3, **A**) and HTelo basket type (PDB: 2mcc, **B**). The results were obtained using Autodock 4.2 and visualised with Chimera.



Struct.	1	2	3	4	5	6	Average
<b>5W77 (A)</b>	1.658	1.805	2.130	2.514	2.790	-	2.18
<b>2MCC (B)</b>	1.832	1.893	1.905	1.920	2.029	2.260	1.97
<b>2MB3 (C)</b>	1.717	2.028	2.038	2.292	2.614	2.788	2.25

Figure S32 – Results from molecular docking studies showing the closest interactions between  $\text{NH}_3$  on compound **1** and guanine oxygens. Distances were measured in Chimera and  $\text{NH}\cdots\text{O}$  distances above 3 Å were excluded. The PDB files listed are c-myc (5w77), HTelo basket type (2mcc) and HTelo hybrid type (2mb3).



Table S2 – Overview of  $\Delta T_m$  ( $^{\circ}\text{C}$ ) values obtained for the FRET competition assay in presence of complex **1** and increasing amounts of CT DNA added to the sample. Three independent experiments were performed and results averaged to determine the standard deviation.

DNA sequence	$\Delta T_m$ ( $^{\circ}\text{C}$ )				
	0 $\mu\text{M}$ CT-DNA	0.6 $\mu\text{M}$ CT-DNA	6.0 $\mu\text{M}$ CT-DNA	60 $\mu\text{M}$ CT-DNA	120 $\mu\text{M}$ CT-DNA
<b><i>HTelo (K)</i></b>	17.4 $\pm$ 0.5	17.3 $\pm$ 0.7	17.0 $\pm$ 0.4	16.7 $\pm$ 0.5	17.0 $\pm$ 0.2
<b><i>HTelo (Na)</i></b>	4.2 $\pm$ 0.5	3.5 $\pm$ 0.4	2.5 $\pm$ 0.7	2.6 $\pm$ 0.7	1.8 $\pm$ 0.6
<b><i>c-myc</i></b>	23.5 $\pm$ 0.1	23.6 $\pm$ 0.3	23.3 $\pm$ 0.4	23.1 $\pm$ 0.1	23.0 $\pm$ 0.3
<b><i>c-kit2</i></b>	13.4 $\pm$ 0.9	13.8 $\pm$ 1.0	13.5 $\pm$ 0.5	12.4 $\pm$ 0.7	11.8 $\pm$ 1.0
<b><i>bcl-2</i></b>	7.4 $\pm$ 0.2	7.2 $\pm$ 0.8	6.6 $\pm$ 0.6	6.1 $\pm$ 0.7	5.9 $\pm$ 0.3